**Comments from the U.S. Department of Interior on the Upper Columbia River Draft Quality Assurance Project Plan for the Phase 2 Sediment Study Appendix D (Table 3 of the DOI comment letter dated May 12, 2011)**

**Comments provided for Teck's information, no response needed**

***Specific Comments on or Related to Appendix D***

| ID | Page | Comment |
|---|---|---|
| 1 | Section A7.4.1; A-10, Lines 26 and 31 | LOE Items 3-7, *C. dilutus* sediment toxicity testing:<br><br>The statement is made that Appendix D provides a detailed evaluation of the relative sensitivity of *C. dilutus* to *H. azteca* or *L. siliquoidea*. This is not correct. No data or evaluations of *C. dilutus* are provided in Appendix D. |
| 2 | Section A7.4.1; A-10, Line 28 | LOE Items 3-7, *L. siliquoidea* sediment toxicity testing:<br><br>The statement is made that Appendix D concludes that *H. azteca* sediment toxicity data would be adequately protective of freshwater mussels. This conclusion is not supported by the data presented in Appendix D. Importantly, key mussel toxicity data are not included in Appendix D and the data that are presented, are presented in a biased manner. |
| 3 | Section A7.4.1; A-10, Line 28 | LOE Items 3-7, *L. siliquoidea* sediment toxicity testing:<br><br>The statement is made that mussel testing should not be conducted because there is no standard sediment toxicity test described for freshwater mussels. This is not adequate rationale for not testing mussels. Dozens of examples are available regarding the use of non-standard methods for USEPA risk assessments. Even within ongoing UCR risk assessment studies, there are dozens of examples of where methods that have not been standardized are being used (e.g., sturgeon water toxicity testing, sturgeon acute toxicity testing, measurement of SEM and AVS, just to highlight a few). |
| 4 | NA | LOE Items 7 and 19, Mussel sediment toxicity testing and press sieving sediment:<br><br>It is good that Appendix D has been provided in the as a "draft", given that there are substantial technical revisions needed to this Appendix as outlined in the comments below. It is also good that Attachment D1 is provided as a "draft final", given this 2008 review is out of date and needs to be updated to include data and findings from studies published in the past 3 years. |
| 5 | D-1 | LOE Item 19, Press sieving Sediment:<br><br>Appendix D is a biased and incomplete representation of the USGS data regarding the influence of sieving sediments on the distribution of metals in two size fractions of sediment (<2 mm versus <0.25 mm). See specific comments below. |
| 6 | D-1 | LOE Item 7, Mussel sediment toxicity:<br><br>Appendix D is a biased and incomplete representation of the USGS data and other published data regarding the acute and chronic sensitivity of mussels to metals in water or in sediment. See specific comments below. |
| 7 | D-1, Line 4 | LOE Item 7, Mussel sediment toxicity: |

| ID | Page | Comment |
| --- | --- | --- |
| | | The correct scientific name is *Lampsilis siliquoidea*, not *L. siliquoidae*. |
| 8 | D-1 | LOE Item 7, Mussel sediment toxicity:<br><br>Appendix D failed to cite and discuss key published literature or presentations associated with the acute and chronic sensitivity of mussels to metals. Instead, Appendix D cites an out of date 2008 review of mussel toxicity data (Appendix A1). Specifically, the following peer review publications and presentations need to be cited and discussed in an updated revision to Attachment D1:<br><br>1) Wang N, Ingersoll CG, Ivey CD, Hardesty DK, May TW, Augspurger T, Roberts AD, van Genderen E, Barnhart MC. 2010. Sensitivity of early life stages of freshwater mussels (Unionidae) to acute and chronic toxicity of lead, cadmium, and zinc in water. *Environ Toxicol Chem* 29:2053-2063. Published August 2010.<br>2) Wang N, Mebane CA, Kunz JL, Ingersoll CG, May TW, Arnold WR, Santore RC, Augspurger T, Dwyer FJ, Barnhart MC. 2009.<br>Evaluation of acute copper toxicity to juvenile freshwater mussels (fatmucket, *Lampsilis siliquoidea*) in natural and reconstituted waters. *Environ Toxicol Chem* 28:2367–2377.<br>3) Wang N, Kunz JL, Ingersoll CG, Brumbaugh WG, Mebane CA, Santore RC, Arnold WR, Gorsuch JW. Influence of dissolved organic carbon on acute and chronic toxicity of copper to rainbow mussel (*Villosa iris*) and the cladoceran (*Ceriodaphnia dubia*). Presented at the 30th meeting of SETAC North America, New Orleans, LA, November 19 to 23, 2009.<br>4) Wang N, Kunz JL, Ingersoll CG, Brumbaugh WG, Mebane CA, Santore RC, Arnold WR, Gorsuch JW. Influence of dissolved organic carbon on acute and chronic toxicity of copper to juvenile freshwater mussels (*Villosa iris*) and cladocerans (*Ceriodaphnia dubia*). Presented at the meeting of SETAC Europe, Seville Spain May 22 to 27, 2010.<br>5) Cope WG, Bringolf RB, Buchwalter DB, Newton TJ, Ingersoll CG, Wang N, Augspurger T, Dwyer FJ, Barnhart MC, Neves RJ, Hammer E. 2008. Differential exposure, duration, and sensitivity of unionoidean bivalve life stages to environmental contaminants. *J N Am. Benthol. Soc.* 27:451-462. |
| 9 | D-1, Line 12 | LOE Item 7, Mussel sediment toxicity:<br><br>Appendix D states sediments for mussel testing must be sieved to <0.25 mm. This is not correct. Sediment testing with mussels can be started with sediment <2 mm (see comments below. |
| 10 | D-1, Line 17 | LOE Item 7, Mussel sediment toxicity:<br><br>Appendix D states that sieving to <0.25 mm enriches total metals and SEM. This is a very biased summary of the data. The <0.25 mm size fraction did not routinely have higher metal concentrations compared to the <2 mm size fraction (see comments below). |
| 11 | D-1 | LOE Item 7, Mussel sediment toxicity:<br><br>It is unfortunate that lines of communication were not permitted to be maintained after the USGS provided Hydroqual with |

| ID | Page | Comment |
|---|---|---|
| | | mussel sediment toxicity data in March 2010. Appendix D has only summarized data for mussel sediment toxicity testing from two studies (Ingersoll *et al.* 2008; Tristate Mining District [TSMD] and Besser *et al.* 2009 Southeastern Missouri [SEMO]). USGS-Columbia has several additional data sets available since the preliminary examples were provided to Hydroqual in March 2010. These include results of (1) Field-collected sediments from sites potentially impacted with coal mining in Virginia and Tennessee, (2) Nickel spiked sediments, and (3) Field-collected sediments contaminated with PCBs. Note in particular the second and third study successfully tested mussels in sediment <2 mm, not in sediment <0.25 mm.<br><br>1) Kunz JL, Wang N, Ingersoll CG, Brumbaugh WG, Kane C, Evans B, Alexander S, Walker C, Bakaletz S, Lott C. Toxicity of coal-associated contaminants in sediment to two freshwater mussels and two commonly tested benthic invertebrates. Presented at the 31st meeting of SETAC North America, Portland OR, November 7 to 11, 2010.<br>2) Besser JM, Brumbaugh WG, Ivey CD, Ingersoll CG. Toxicity of nickel to benthic invertebrates: Nickel bioavailability and species sensitivity in freshwater sediments. Presented at the 31st meeting of SETAC North America, Portland OR, November 7 to 11, 2010. |
| 12 | D-2, Line 3 | <u>LOE Item 7, Mussel sediment toxicity</u>:<br><br>Appendix D states that in one study (Besser *et al.* 2009) the sediments were marginally more toxic to mussels compared to amphipods. This is not a correct statement. The sediments were frequently more toxic to mussels with an 80% correspondence between laboratory and field effects on mussels, in relation to metals chemistry of the sediments. Moreover, Appendix D failed to report the conclusion from Besser *et al.* (2009) regarding the lower sensitivity of mussels compared to amphipods in the TSMD project. Specifically, Besser *et al.* (2009) suggested that the older and mussels (3- to 4-month old mussels) tested in the TSMD project (Ingersoll *et al.* 2008) may have been less sensitive compared to the younger mussels tested in the SEMO project (2-month old mussels; Besser *et al.* 2009). |
| 13 | D-2, Line 14 | <u>LOE Item 19, Press sieving sediment</u>:<br><br>Sediments are not wet sieved, the sediments are press sieved. |
| 14 | D-2, Line 22 | <u>LOE Item 7, Mussel sediment toxicity:</u><br><br>Appendix D states that when water quality and DOC are considered, that mussels are not among the most sensitive species to metals. Support for this statement is referenced in Wang *et al.* (2009). This is a mis-representation of the acute data summarized in Wang *et al.* (2009; a paper co-authored by Hydroqual). Specifically, see Figure 4 in Wang *et al.* (2009; also provided as supplemental technical information by Appendix D). Based on the USEPA (2007) BLM for copper, Wang *et al.* (2009) concluded: "Five of the six genus mean acute values (GMAVs) for mussels were within the range of GMAVs for the ten most sensitive genera used to derive the U.S. EPA acute WQC for copper (Fig. 4)." Furthermore, Wang *et al.* (2009) concluded that "Nearly 70% of USEPA (2007) copper BLM-normalized EC50s for fatmucket tested in natural waters were below the final acute value used to derive the U.S. Environmental Protection Agency acute WQC for copper, indicating that the criterion might not be protective of fatmucket and perhaps other mussel species." Additionally, Wang *et al.* (2009) concluded: "The species mean acute value (SMAV) for fatmucket would rank this mussel species |

| ID | Page | Comment |
|---|---|---|
| | | among the four most sensitive species used in the derivation of the U.S. EPA acute WQC for copper (USEPA 2007 BLM WQC)." Wang *et al.* (2010; presentations co-authored by Hydroqual) concluded that: "The USEPA (2007) copper BLM-normalized acute EC50s and chronic EC20s for the mussel *Villosa iris* and the chronic EC20s for the cladoceran *Ceriodaphnia dubia* in waters with the DOC concentrations of 2.5 to 10 mg C/L were equal to or less than the final acute value and final chronic value in the BLM-based WQC for copper , indicating that the copper WQC might not adequately protect the *V. iris* from acute and chronic exposure, and the cladoceran from chronic exposure."<br><br>Hence, the statement made by Appendix D that when water quality and DOC are considered, mussels are not among the most sensitive species to copper, is clearly not supported by the data presented in Wang *et al.* (2009, coauthored by Hydroqual). |
| 15 | D-2, Line 22 | LOE Item 7, Mussel sediment toxicity:<br><br>Appendix D states that when water quality and DOC are considered, that mussels are not among the most sensitive species to metals. This statement is not correct.<br><br>Wang *et al.* (2010) concluded that "Chronic water toxicity values for the mussel *Lampsilis siliquoidea* (fatmucket) were 10 µg Pb/L, 6.0 µg Cd/L, and 63 and 68 µg Zn/L. ). When *L. siliquoidea* toxicity data from the present study and the literature were included in updated databases for deriving U.S. Environmental Protection Agency water quality criteria, mussel genus mean acute values were in the lower percentiles of the sensitivity distribution of all freshwater species for lead (the 26th percentile), cadmium (the 15th to 29th percentile) or zinc (the 12th to 21st percentile). Mussel genus mean chronic value was the lowest value ever reported for lead (the 9th percentile), but was near the middle of the sensitivity distribution for cadmium (the 61st percentile) or zinc (the 44th percentile). These results indicate that mussels were relatively sensitive to the acute toxicity of these three metals and to the chronic toxicity of lead, but were moderately sensitive to the chronic toxicity of cadmium or zinc compared to other freshwater species."<br><br>Furthermore, Wang *et al.* (2010) concludes "zinc EC50s and NOECs for juvenile mussels listed in Wang *et al.* (2010) were normalized using the BLM, which was similar to the approach used to derive the 2007 U.S. EPA WQC for copper (USEPA 2007). The BLM-normalized zinc GMAVs for mussels were between the 15th and 21th percentiles in the sensitivity distribution (Fig. 2A), which was similar to the ranks based on hardness normalized GMAVs (the 12th to 21st percentile; Fig. 1C). The five BLM normalized GMAVs for mussels were still among the most sensitive genera of freshwater species, but all were >2-fold above the BLM-derived FAV (Fig. 2A). The BLM-normalized zinc NOEC for fatmucket was at the 33rd percentile (Fig. 2B), which was slightly lower than the rank based on the hardness-normalized GMCV (the 44th percentile; Fig. 1F), but was 3.9-fold above the BLM-derived CCC. Therefore, the proposed draft BLM-derived acute and chronic WQC for zinc (Fig. 2) would be more protective of freshwater mussels than the current or revised hardness-dependent WQC for zinc (Figs. 1C and 1F)." |
| 16 | D-3, Line 14 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Whole-sediment toxicity tests with mussels are conducted for 28 days, not 21 days (as reported by Besser *et al.* 2009, |

| ID | Page | Comment |
|---|---|---|
| | | Ingersoll *et al.* 2008, MacDonald *et al.* 2009). |
| 17 | D-3, Line 18 | LOE Item 19, Press sieving sediment:<br><br>Appendix D states that "it is not unusual for materials larger than 2 mm to be removed prior to measuring sediment chemistry, and so it would not typically be expected that sediments sieved to 2 mm would have chemical characteristics that are appreciably different from bulk sediments." We agree with the statement that sieving sediments to 2 mm would not be problematic in sediment toxicity testing in relation to sediment chemistry. |
| 18 | D-3, Line 24 | LOE Item 19, Press sieving sediment:<br><br>Appendix D states metal contaminated sediments are typically anoxic. This is not a correct statement. At sites contaminated primarily by metals (including the UCR), AVS, tends to be relatively low (e.g., Ingersoll *et al.*2008, Besser *et al.* 2009, MacDonald *et al.* 2009, USEPA 2005 [UCR], Besser *et al.* 2008; Besser JM, Brumbaugh WG, Ivey CD, Ingersoll CG, Moran PW. 2008. Biological and chemical characterization of metal bioavailability in sediments from Lake Roosevelt, Columbia River, Washington USA. Arch Environ Toxicol. Chem.: 54:557-570). Moreover, Appendix D concluded that sieving sediment did not influence concentrations of AVS in the TSMD study (Figure 5). |
| 19 | Figures 1, 2, 3, and 4 | LOE Item 19, Press sieving sediment:<br><br>Appendix D attempts to demonstrate was substantial metal enrichment in the <0.25 mm sediment compared to the <2 mm sediment in the (1) TSMD sediments (Figure 1 and 3) and in SEMO sediments (Figures 2 and 4). Appendix D concludes from these plots that there was a 5-fold enrichment of copper, 7-fold enrichment of nickel, 10-fold enrichment of zinc, and a 12- fold enrichment of lead (Figure 3) and there was a 2.5-fold enrichment of zinc, 5-fold enrichment of cadmium, a 5-fold enrichment of lead, a 4-fold enrichment of copper, and a 3 fold enrichment of nickel (Figure 4). Appendix D is a biased and incomplete summary of the data (highlighting only the extremes of the upper distribution). Specifically Figures 3 and 4 illustrate that the majority of the sediments exhibited enrichment factors less than 2, with a substantial number of samples with higher enrichment of metals in the 2 mm fraction compared to the 0.25 mm fraction. Regardless of this bias presentation of these data, mussel sediment toxicity testing does not require sieving to size fractions <2 mm. |
| 20 | Figures 5,6,and 7 | LOE Item 19, Press sieving sediment:<br><br>In Figure 5, Appendix D attempts to demonstrate that concentrations of AVS and SEM metals are substantially enriched in the 0.25-mm size fraction compared to the 2-mm size fraction of sediment. These plots need to include a 1:2 and 2:1 line (as is typically the practice for BLM modeling by Hydroqual; e.g., see Figures 3 or 4 in Wang *et al.* 2009). Importantly, the data presented in Figures 5, 6, and 7 do not support the premise made by<br>Appendix D that AVS or SEM are substantially enriched in the 0.25-mm size fraction compared to the 2-mm size fraction (erroneously stated in the text as the 0.2-mm size fraction rather than the 2-mm size fraction). |
| 21 | D-14, Line 3 | LOE Item 19, Press sieving sediment:<br><br>Appendix D states that sediment sieving is generally conducted only if it is appropriate or necessary prior to toxicity testing |

| ID | Page | Comment |
|---|---|---|
| | | to removed coarse materials. We agree with the statement as has been recommended in the sediment LOE to press sieve sediment to 2 mm. |
| 22 | Figure 8 | LOE Item 19, Press sieving sediment:<br><br>Appendix D provides convincing data as to why sediments should be consistently sieved to 2 mm across the entire UCR (Figure 8). |
| 23 | D-14, Line 31 | LOE Item 19, Press sieving sediment:<br><br>Appendix D concludes that ammonia would be elevated in sieved sediment. This conclusion has not been adequately documented in Appendix D. |
| 24 | D-14, Line 15 | LOE Item 19, Press sieving sediment:<br><br>Burton *et al.* (2003) does not provide standard methods for sieving. Moreover, testing sediments as soon as possible is not recommended in actual standard methods (e.g., ASTM E1706, USEPA 2000). |
| 25 | D-16, Line 1 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Attachment D1 provides a 2008 review of mussel toxicity data to conclude that mussels are marginally sensitive to many metals and ammonia. The review provided in Attachment D1 is out of date. This 2008 review iin Attachment A needs to be updated based on studies published over the past 3 years (see comments above). |
| 26 | Figures 9, 10, and 11 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Appendix D provides a bias and incomplete summary of the relationship between sediment chemistry and sediment toxicity in the TSMD study (Figure 9, 10, 11). The discussion of relationships between reported toxicity and chemistry provide in MacDonald *et al.* (2009) should be cited and discussed rather than use an arbitrary toxicity threshold of 80% survival as the only endpoint evaluated. Importantly, growth of mussels in sediment exposures has been demonstrated to be a sensitive endpoint (at responses less than 20%; e.g., Besser *et al.* 2009, Wang *et al.* 2010). Figure 11 does not demonstrate that amphipods were "clearly more sensitive than mussels." |
| 27 | Figure 12 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Appendix D provides a very biased summary of the relationship between sediment chemistry and sediment toxicity in the SEMO study (Figure 12). Sediments were not just "apparently" more toxic to mussels than to amphipods, the sediments with elevated metals "were" more toxic to mussels (as described by Besser *et al.* 2009). Importantly, the least sensitive endpoint (survival) was presented from Besser *et al.* (2009) in an apparent to attempt to dampen the highly sensitive toxicity endpoint of mussel growth in the SEMO study. |
| 28 | D-21, Line 1 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Appendix D concluded that mussels may respond to the physical characteristics of the sediments. No data are presented |

| ID | Page | Comment |
|---|---|---|
| | | in Appendix D to support the assertion that mussels are sensitive to physical characteristics of sediments. Moreover, the sediment LOE requires testing of reference sediments in order to evaluate potential influence of physical characteristics of sediments on not only mussels, but on amphipods or midge too (see Item 18 in Table 1). |
| 29 | D-21, Line 8 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Appendix D concludes that effects on mussels were not related to metal concentrations in the SEMO sediment. This statement is not correct. Growth response of mussels was highly correlated to metal concentrations in the sediment samples. Moreover, elevated metals in the SEMO sediments correctly classified all of the sediments with reduced growth of mussels (Besser *et al.* 2009). |
| 30 | D-22, Line 3 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Appendix D references a personal communication to CG Ingersoll that additional study is needed to determine if mussels can be recovered from coarser sediment. It is unfortunate that the authors of Appendix D were not permitted to consult with USGS-Columbia after the TSMD data and the SEMO data were provided to Hydroqual in March 2010. Specifically, there are additional datasets available since March 2010: (1) Field-collected sediments from sites potentially impacted with coal mining in Virginia and Tennessee, (2) Nickel spiked sediments, and (3) Field-collected sediments contaminated with PCBs. Note in particular the second and third study successfully tested mussels in sediment <2 mm, not in sediment <0.25 mm. |
| 31 | D-23, Line 6 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Appendix D concluded that the mussel *Anodonta* is much less sensitive that standard toxicity testing organisms. We agree that less sensitive species from this less sensitive genera of *Anodonta* should not be used in testing UCR sediments. Moreover, a sensitive representative genera (such as *Lampsilis* or *Villosa*) should be tested. Importantly, testing of resident species is not required in standard sediment methods (e.g., *Hyalella azteca*; ASTM E1706, USEPA 2000). Importantly, Appendix D has not provided convincing data to conclude that 28-day whole sediment testing *H. azteca* (much less 10-day exposures as proposed in the sediment QAPP) would provide adequate protection to mussels inhabiting sediments of the UCR. This is the reason that the sediment LOE recommends that mussel responses be compared to other commonly tested species in whole-sediment toxicity tests with UCR sediments (Item 7 in Table 1 of the DOI comments). |
| 32 | Attachment D1 | LOE Item 7, Mussel sediment toxicity testing:<br><br>It is good that Attachment D1 is provided as a "draft final", given this 2008 review is out of date and needs to be updated to include data and findings from studies published in the past 3 years (see comments above). It is surprising that this 2008 report states that the review was prepared to address the recommendation of mussel sediment test in the UCR (a year before mussel sediment testing was ever described in the initial sediment LOE in 2009). |
| 33 | Attachment D1, page ix | LOE Item 7, Mussel sediment toxicity testing:<br><br>Attachment D1 concludes there are significant limitations on the use of mussels as biomonitoring organisms; however, no |

| ID | Page | Comment |
|---|---|---|
| | | studies are cited to support this conclusion. |
| 34 | Attachment D1, Section 2.1 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Attachment D1 concludes that mussels are only available for testing during the season they are reproductively mature. This statement is not correct. See guidance presented in ASTM E2455 regarding the year-round availability of glochidia for propagating juvenile mussels for toxicity testing. |
| 35 | Attachment D1, Section 2.1 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Attachment D1 reports that juvenile mussels transformed *in vitro* can be used for toxicity testing. This is not correct. ASTM E2455 requires that toxicity tests not be conducted with *in vitro* transformed juveniles. |
| 36 | Attachment D1, page 2-3, 2nd and 3rd paragraph | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Cite and discuss the review paper by Cope *et al.* (2008) regarding exposure duration. Ingersoll *et al.* (2006) is incorrectly cited (not 2007). Discuss chronic juvenile mussel methods (e.g., 21- to 28-d exposures) not just 96-h exposures. |
| 37 | Attachment D1, Section 2.4.2 and Section 3.2.2.1 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Wang *et al.* (2010; chronic DOC copper toxicity to mussels and amphipods) addresses equilibration of copper in diluters (minimal impact on the toxicity to organisms with a 24-h equilibration period). Moreover, USEPA WQC for metals include nearly exclusively toxicity data generated from diluters that do not have a 24-h equilibration period. |
| 38 | Attachment D1, Section 2.4.3 and Section 3.2.1.2 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Cite and discuss the findings of Wang *et al.* (2009, 2010) regarding the BLM modeling of copper or zinc toxicity to mussels in acute or chronic exposures. |
| 39 | Attachment D1, Section 3.2, Table 3-1 and Figure 3-1 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Cite and discuss the findings of Wang *et al.* (2009, 2010) regarding acute or chronic toxicity testing of mussels with copper, cadmium, zinc, or lead. |
| 40 | Attachment D1, Section 3.2.1.1, | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>Cite and discuss the findings of Wang *et al.* (2010) regarding the lack of sensitivity of glochidia relative to juvenile mussels exposed to zinc, cadmium, or lead. |

| ID | Page | Comment |
|---|---|---|
| | page 3-8 and Figures 3-3 and 3-4 | |
| 41 | Attachment D1, Section 3.2.2 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Cite and discuss the findings of Wang *et al.* (2009, 2010) regarding chronic toxicity testing of mussels with copper, cadmium, zinc, or lead. |
| 42 | Attachment D1, Figure 3-6 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Expand this figure to include chronic toxicity testing of mussels with copper, cadmium, zinc, or lead (Wang *et al.* 2009, 2010). |
| 43 | Attachment D1, Figure 3-7 | LOE Item 7, Mussel sediment toxicity testing:<br><br>The ACRs cited in Figure 3-7 (from Wang *et al.* 2007c) are not correct. Moreover, include the findings of Wang *et al.* (2009, 2010) regarding chronic toxicity testing of mussels with copper, cadmium, zinc, or lead. |
| 44 | Attachment D1, Section 4 | LOE Item 7, Mussel sediment toxicity testing:<br><br>The acute metal toxicity data summarized in Figures 4-1, 4-2, 4-3, 4-4, and 4-6 (and the associated text) needs to be updated to include the findings of Wang *et al.* (2009, 2010) regarding acute or chronic toxicity testing of mussels with copper, cadmium, zinc, or lead. |
| 45 | Attachment A, page 4-2, 2nd paragraph | LOE Item 7, Mussel sediment toxicity testing:<br><br>Attachment D1 concludes that the differences in age between mussels tested in Wang *et al.* (2007b) confound the findings compared other commonly tested species. This conclusion is not correct. Consistent life stages (newly transformed or about 2-months post transformation were tested relative to commonly tested species). Moreover, this discussion needs to be expanded to include a summary of the findings of Wang *et al.* (2009, 2010) regarding chronic toxicity testing of mussels with copper, cadmium, zinc, or lead. |
| 46 | Attachment D1, Section 4, page 4-10 | LOE Item 7, Mussel sediment toxicity testing:<br><br>The acute ammonia toxicity data summarized in Figure 4-7 (and the associated text) needs to be updated to include the findings of Wang *et al.*(2009, 2010) and Miao *et al.* (2010) regarding acute and chronic toxicity testing of mussels with ammonia.<br>1) Wang N, Erickson RJ, Ingersoll CG, Ivey CD, Brunson EL, Augspurger T, Barnhart MC. 2008. Influence of pH on the acute toxicity of ammonia to juvenile freshwater mussels (Fatmucket, *Lampsilis siliquoidea*). *Environ Toxicol Chem* 27:1141-1146.<br>2) Miao J, Barnhart MC, Brunson EL, Hardesty DK, Ingersoll CG, Wang N. 2010. An evaluation of the influence of substrate on the response of juvenile freshwater mussels (fatmucket, *Lampsilis siliquoidea*) in acute water exposures |

| ID | Page | Comment |
|---|---|---|
| | | to ammonia. *Environ Toxicol Chem* 29:2112-2116.<br>3) Wang N, Ingersoll, CG, Miao J, Brunson EL, Hardesty DK, Consbrock RA, Barnhart MC. 2010. Evaluation of the influence of sediment on the response of juvenile mussels (Lampsilis siliquoidea) in acute and chronic water exposures to ammonia. Presented at the 31st meeting of SETAC North America, Portland OR, November 7 to 11, 2010. |
| 47 | Attachment D1, Section 4, page 5-4 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>The discussion of the quality of the data for glochidia toxicity tests is not clearly described. ASTM E2455 provides guidance as to what constitutes an acceptable glochidia toxicity test. It is not clear from this write up if these requirements were met in this data compilation. |
| 48 | Attachment D1, Table 6-1 | LOE Item 7, Mussel sediment toxicity testing:<br><br>Table 6-1 cited from a USGS quarterly report needs to be update based on methods reported in Ingersoll *et al.* (2008), Besser *et al.* (2009), Wang *et al.*(2010), and Kunz *et al.* (2010). |
| 49 | Attachment D1, Section 6.1 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>We agree with the conclusion in Attachment D1 that ASTM standard sediment toxicity testing methods for other invertebrates (ASTM E1706) can be modified to conduct whole sediment toxicity tests with mussels. See Ingersoll *et al.* (2008), Besser *et al.* (2009), Wang *et al.* (2010), and Kunz *et al.* (2010) for details. |
| 50 | Attachment D1, Section 6.1 | <u>LOE Item 7, Mussel sediment toxicity testing</u>:<br><br>We agree with the conclusion in Attachment D1 that testing of mussels should be done with taxonomically similar species. This is why mussel testing has been recommended in the sediment LOE to evaluate risks of contaminated sediments on mussels inhabiting the UCR (Item 7 in Table 1), rather than depend on the use of other surrogates such as amphipods (Items 4 and 6 in Table 1) or midge (Items 3 and 5 in Table 1). Moreover, Wang *et al.* (2007a,b,c; 2008, 2009, 2010) have demonstrated that there are relatively consistent responses across various species or genera of mussels in acute or chronic exposures to metals or ammonia. Hence, use of a surrogate from the genera of *Lampsilis* or from *Villosa* would likely provide a representative measurement of risk of metal-contaminated sediments to mussel species currently inhabiting or historically inhabiting the UCR. |